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Evaluation of antioxidant activity and texture profile of tender-young and king coconut (*Cocos nucifera*) mesocarp

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Abstract

The present study is aimed at analyzing the antioxidant activity and texture profile of mesocarps of two varieties-tender king and young coconut. Antioxidant activity was determined using the free radical 2, 2-diphenyl- 1-picrylhydrazyl (DPPH) method. The texture was analyzed using the Texture profile analysis test. The antioxidant activity (IC_{50} value) of tender mesocarp of king and young coconut were 360.1 ± 26.4 mg/L and 448.4 ± 34.6 mg/L respectively and which was significantly different to each other ($p < 0.05$). But among the mesocarps of tender king and young coconut, there is no significant difference in texture parameter "hardness" however; there was a significant difference in texture parameter "chewiness" ($p < 0.05$). The hardness and chewiness values of fresh king and young coconut mesocarps were (1306.3 ± 44.6), (24.92 ± 2.06) and (1281.3 ± 70.8) and (79.62 ± 5.56) respectively. Since, the tender king coconut mesocarp having higher antioxidant activity and lower chewiness than young coconut, there is a sound activity to develop a healthy food product from tender mesocarp of king coconut.

Keywords: antioxidant activity, *Cocos nucifera*, king coconut, mesocarp, young coconut

1. Introduction

Cocos nucifera (L), member of family Arecaceae is commonly called the "coconut tree" and is the most naturally wide spread fruit plant on earth [1]. This plant is originally from South East Asia [2]. The coconut is the fruit of the coconut tree, a drupe containing prized flesh and water (endosperm) protected by three distinct sections such as the exocarp, mesocarp, and endocarp [3]. The epicarp, which is the outermost skin of the fruit and the mesocarp which is the heavy, fibrous and tanned when dry, has many industrial uses. The endocarp is the inner hard dark core [4]. There are two main varieties of tender coconuts used for getting coconut water. They are young coconut which is green in colour and king coconut which is light orange in colour. According to Jarimopas & Ruttanadat (2007) [5] and Gatchalian *et al.*, (1994) [6], young coconut is highly nutritious and is one of the most popular export fruits from Southeast Asian countries.

Antioxidant activity is predominated in constituents of endocarp and coconut water [7]. Coconut has been used for the preparation of many foods and beverages other than oil extraction. Coconut testa is the brown coloured outer thin layer covering the coconut kernel. It was identified that the coconut testa contains high antioxidant and phytochemicals. The antioxidants identified are phenolic acids, flavonoids and total tocopherol [8]. Antioxidants are the radical scavengers which help in delaying or prevention of oxidation by trapping the free radicals [9]. Many parts of *Cocos nucifera* have proven to contain phenolic compounds and flavonoid that support antioxidant activity.

There are plenty of studies on the availability of antioxidant present in coconut oil, coconut water and the kernel. But limited studies are available on the antioxidant activity mesocarp, which is the part between skin and the kernel. This study aims to analyze the antioxidant activity of tender coconut mesocarp both, fresh and treated and create a path towards developing an edible product from the mesocarp.

2. Materials and Methodology

2.1 Study location

The research was mainly carried out at the laboratory of the department of Food Science and Technology, Faculty of Applied sciences, University of Sri Jayewardenepura.

2.2 Sample preparation

The tender coconut fruits of the two varieties young coconut and king coconut were taken and each of the coconut fruit is halved using a sharp

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curved knife while removing the coconut water. Then the mesocarp of the tender fruit was cut in to small pieces using a sharp stainless steel knife. These mesocarp pieces were used for the subsequent use of the study.

2.3 Sample extraction

For sample extraction, the procedure described by Appaiah *et al.*, (2016) [8] was followed with some modifications. Initially the mesocarp sample was ground using the mortar and pestle into a paste. Thereafter, 10.0000g ± 0.0001g of ground mesocarp was weighed using the analytical balance PA214. Then the weighed sample was added into the thimble and covered with cotton wool and placed in the soxhlet apparatus. 250ml of 95% ethanol was added the round bottom flask and the extraction was carried out for 3 hours at 78.3°C. The extracted solvent was evaporated under vacuum at 40-45°C using a rotary evaporator, to get the gummy concentrate of reddish colour.

2.4 Analysis of the properties of coconut mesocarp

2.4.1 Analysis of antioxidant activity

Sample stock solution

Sample stock solutions having concentration of 0.01g/ml (10mg/ml) were prepared in a 100ml volumetric flask by dissolving 1.0000 ± 0.0001g of red gummy extract of each extraction with methanol.

From each of the solution prepared, a dilution series was made by mixing 1 part of the sample stock solution to 4 parts of absolute methanol. From this dilution six different concentrations of sample solution were prepared, having concentration of 2000mg/L, 400.0mg/L, 80.0mg/L, 16.0mg/L, 3.2mg/L and 0.64mg/L respectively.

DPPH (1, 1-diphenyl-picryl hydrazyl) radical scavenging activity

For the DPPH assay, the procedure of Brand-Williams *et al.*, (1995) [10] with some modifications was followed.

a. Preparation of DPPH stock solution

Using the analytical balance 0.0039g ± 0.0001g (3.9mg) of DPPH chemical was weighed and the stock DPPH solution of 10⁻⁴ M concentration was prepared by dissolving the measured quantity (3.9 mg) of DPPH in 100ml of absolute methanol to obtain an absorbance of 0.900 ± 0.02 units at a wavelength of 517nm, using U.V-Vis spectrophotometer (UV mini -1240 model).

*After preparing the DPPH solution, it was kept in dark room for 30minutes and then the absorbance was measured.

b. Determination of antioxidant activity

A blank sample was prepared by mixing 0.5ml of methanol and 2.5ml of absolute methanol. A Control sample was prepared by mixing 2.5ml of 10⁻⁴ M DPPH solution and 0.5ml of absolute methanol. From each dilution series of the sample (for each of the six concentrations of the mesocarp extract), 0.5ml of sample solution was taken and mixed with 2.5ml 10⁻⁴M DPPH stock solution. For all these volume measurements micropipette was used.

These mixtures were kept in dark place at room temperature for 15 minutes, then the resulting solution was vortexed for 30 seconds and the absorbance was read at 517nm in U.V-visible spectrophotometer. Thereafter the percentage inhibition of absorbance was calculated for each dilution using the following equation.

$$\% \text{ inhibition} = \left[\frac{A_{\text{control}} - A_{\text{sample}}}{A_{\text{control}}} \right] \times 100$$

Where;

A control = absorbance value of the DPPH solution of the control sample.

A sample = absorbance value of the DPPH solution of the coconut mesocarp extract sample.

The calculated percentage of inhibition at 517nm was plotted as a function of concentration of the coconut mesocarp extract sample. Then the concentration of sample, which gives 50% inhibition activity was estimated as the IC₅₀ value from regression analysis, using the software MINITAB®17.

For each concentration of the dilution series of each coconut mesocarp extract, the measurement of absorbance at 517nm was triplicated. As the sample (coconut mesocarp) is triplicated and the measurement is also triplicated, there will be nine absorbance values, inhibition% and IC₅₀ values for each concentration of coconut mesocarp extract.

c. Drawing the standard curve

Gallic acid was used as the standard for DPPH Assay. Six different concentrations of Gallic acid such as 1, 2, 3, 4, 5, 6 mg/L, were prepared using absolute methanol as the solvent. From each concentration of Gallic acid solution, 0.5ml was pipetted into a tube using the micropipette and mixed with 2.5ml of 10⁻⁴M DPPH solution. Thereafter the absorbance of the resulting solution was measured in UV-Vis spectrophotometer at 517nm, after keeping the mixed solution in a dark place at room temperature for 15 minutes.

The percentage of inhibition was calculated for each concentration of Gallic acid solution prepared. The percentage inhibition values were plotted as a function of concentration of standard antioxidant (Gallic acid). This will impart the standard curve. The concentration of Gallic acid solution which gives 50% inhibition activity was estimated as the IC₅₀ value for standard reference from regression analysis using the software MINITAB®17.

Comparison of antioxidant activity in fresh mesocarps of two coconut varieties

The IC₅₀ values obtained from the graph of percentage of inhibition vs. concentration of test solution in the DPPH assay of fresh mesocarp extract were compared using the 2-independent sample t-test method at 95% confidence level using the MINITAB®17 software.

2.5.2 Analysis of texture profile of coconut mesocarps

In the analysis of texture profile, the procedure described by Thomson *et al.*, (1992) [11], was followed with some modifications.

Texture profile analysis (TPA) was performed with a view to differentiate the mesocarp of the two coconut varieties under different treatments. Optimum instrumental test conditions set were 4mm (T44), pre-test speed 2.00mm/s, test speed of 1.00mm/s and post-test speed of 3.00mm/s. The target parameter is the deformation of 75%. The load was 1000g (10N). The TPA test is a two cycle test.

After setting all the parameters in the texture Pro Ct software of the Brook field CT3 texture analyzer, the test was run initially to locate the base. After that each coconut mesocarp slices having approximately 7mm thickness, was placed on the sample table of the texture profile analyzer and the load needed to make a 75% deformation in the mesocarp slice was

determined by running the test and drawing a graph on load vs. time using the texture Pro Ct software. Then the parameters needed for the analysis were selected and the results were recorded. The values of parameter such as hardness cycle-1, hardness cycle-2, and chewiness were recorded and analyzed.

The samples were triplicate and for each replicate five measurements were taken. Thus, each treated and fresh mesocarp of each coconut variety had 15 readings. Thereafter, the mean of the texture parameter values are obtained with standard deviation, using descriptive statistics of MINITAB®17 software. These parameters were also analyzed using ANOVA at 5% significant level followed by a comparison using Tukey test by statistical software MINITAB®17. This was done to test whether there is a significant difference of texture parameters among the different treatments of each variety of coconut.

2.6 Data analysis

2.6.1 Statistical analysis

All analysis was carried out in triplicates and was reported as mean \pm standard deviation. The collected data was finally analyzed by using MINITAB®17 software. For the parametric data analysis, One-way ANOVA was used at 95% confidence interval and for the pair wise comparison of the means

Tukey's Analysis was used. For the comparison of two samples, two-independent sample t-test was used.

2.6.2 Parametric analysis

Two independent sample t-test was used to compare the two samples, to determine significant difference ($P < 0.05$) between the mean values of the analyzed parameters.

3. Results and Discussion

3.1 Analysis of antioxidant activity of fresh young coconut and king coconut

The activity of the natural antioxidant to reduce the DPPH free radical will be measured by decrease in the absorbance of U.V visible spectrum at 517nm wave length, and the Gallic acid which is a good antioxidant was used as the positive control and reference to the IC₅₀ values of the sample.

IC₅₀ value is the antioxidant concentration of the sample that shows 50% inhibition activity of DPPH free radicals and it is indicated in mg/L. So if lesser concentration of the sample is needed for half (50%) maximum inhibitory action, the sample has higher antioxidant activity, meaning that IC₅₀ value is inversely proportional to the antioxidant activity.

The IC₅₀ values of the DPPH assay done for the fresh tender young coconut mesocarp and king coconut mesocarp and the standard Gallic acid which was used as the positive control are given in the table 1 along with their standard deviations.

Table 1: DPPH radical scavenging assay based on IC₅₀ value (mg/L) in the mesocarp of tender young coconut and king coconut

Property	King coconut	Young coconut	Standard antioxidant
IC ₅₀ value (mg/L)	360.1 \pm 26.4	448.4 \pm 34.6	4.3180 \pm 0.0117

*Data presented as mean values for triplicates with triplicate measurements in each replicate \pm S.D (n=9).

The IC₅₀ value of king coconut and young coconut given in the table 1 were 360.1 and 448.4 mg/L respectively and that indicate 360.1 and 448.4mg/L of king coconut and young coconut mesocarp extracts are required to scavenge 50% free radicals in 10⁻⁴M DPPH solution.

According to the results, king coconut had lower IC₅₀ value than that of young coconut. It indicates that king coconut had higher antioxidant activity than the young coconut due to low IC₅₀ values which indicate high antioxidant activity.

Coconut testa is the brown coloured outer thin layer covering the coconut kernel. It was identified as an important part of tender coconut because; it contains that high antioxidant and phytochemicals. The identified antioxidants are phenolic acids, flavonoids and total tocopherol. The IC₅₀ value of coconut testa extracts from previous study was 0.06- 0.5 mg/ml which is equal to 60-500 mg/L [8]. Thus, the IC₅₀ value of coconut mesocarp obtained in this study was very closer to IC₅₀ value of coconut testa.

According to Wasala *et al.*, (2015) [12], among the fruits bael fruit has the highest antioxidant activity. The IC₅₀ value of antioxidant activity of bael fruit is 103.89 mg/L. The current study indicates that king coconut mesocarp has antioxidant activity of about 1/3rd of bael fruit and young coconut mesocarp has about 1/4th of bael fruit.

However when comparing the IC₅₀ values of the two varieties of coconut against chemical antioxidant Gallic acid (4.3180 \pm 0.1171), they are lower in antioxidant activity as they had higher IC₅₀ values than Gallic acid. Hence, synthetic antioxidants have higher performance level than the natural ones because the natural antioxidants show a greater reluctance when donating hydrogen atoms in order to prevent oxidation [13].

Based on the 2-independent sample t-test, the difference among the mean IC₅₀ values obtained for king coconut and young coconut is significantly different at 95% confidence level.

However when considering about this assay, DPPH is a stable nitrogen-radical that bears no similarity to the highly reactive and transient peroxy radicals involved in lipid peroxidation. Many antioxidants that react quickly with peroxy radicals may react slowly or may even be inert to DPPH due to steric inaccessibility [14]. Interpretation is complicated when the test compounds have spectra that overlap DPPH at 515 nm. Carotenoids, in particular, interfere [10].

Some authors have reported that DPPH reactions should be tested in multiple solvents, at a minimum MeOH (not EtOH, which forms reactive radicals that may interfere with the assay). It was noted that compounds active in this DPPH reaction are glutathione, aromatic amines (p-phenylene diamine and p-aminophenol), and a-tocopherol (Vitamin E - 2:1 stoichiometry) and polyhydroxy aromatic compounds (hydroquinone and pyrogallol). On the other hand, monohydric phenols (tyrosine), simple sugars (glucose), purines and pyrimidines, do not react, while proteins are precipitated. It was also noted that "inorganic ions in lower valence states may of course interfere and must be eliminated or determined separately" which presumably applies most importantly to ferrous iron [15].

According to Sultana *et al.*, (2007) [16], antioxidant retention depends the type of vegetable, bioavailability of phenolic, temperature, location in vegetables cutting the synergic activity of the structure and on the antioxidant system assayed. In accordance with previous study this assay supports the fact antioxidant activity of coconut mesocarp depends on the variety of coconut.

3.2 Texture profile analysis of fresh, tender king coconut and young coconut mesocarp.

In this study, the textural properties such as Hardness and Chewiness were analyzed. Hardness is the force required to make a certain deformation in the product. In this analysis, texture profile had two cycles (Cycle 1, deformation at 1st cycle and cycle 2, deformation at 2nd cycle due to hardness). Chewiness measured in mJ is the energy required to chew the

product during the mastication process. These parameters have an effect on the sensory attributes of the product.

The mean hardness values of cycle-1 and cycle-2 and the mean chewiness values of the texture profile analysis test and the p-values of 2-independent sample t-test done for fresh-tender coconut mesocarps of king and young coconut are given in table 2.

Table 2: Mean hardness and chewiness parameter values for fresh king coconut and young coconut mesocarp.

Property	Hardness cycle-1(g)	Hardness cycle-2(g)	Chewiness (mJ)
King coconut	1306.3 ± 44.6	884.0 ± 25.2	24.92 ± 2.06
Young coconut	1281.3 ± 70.8	912.7 ± 58.2	79.62 ± 5.56
P-value	0.26	0.09	0.00

*Data a presented as mean values for the triplicates with five measurements in each replicate ±SD (n=15)

According to the table 2, there is no significant difference between, in the hardness of fresh-tender king coconut and young coconut mesocarp at cycle-1 (p<0.05). The P-value of hardness cycle -2 (0.091) is also is greater than that of default $\alpha = 0.05$. Hence, there is no significant difference between two types of coconut in the hardness of mesocarp at cycle-2 too.

However for both king and young coconuts, the hardness at cycle -1 is greater than the hardness at cycle -2 because the hardness at cycle -2, measures the hardness of the product deformed by the cycle-1 of the test.

At 5% significant level, the P-value of chewiness (0.000) is less than that of default α (0.05). Hence there is a significant difference between the chewiness of fresh tender king coconut and young coconut mesocarp. According to the mean values, tender king coconut mesocarp had lesser chewiness values (24.92mJ) than tender young coconut (chewiness-79.62 mJ).

The composition of the raw material has a major role to play in the textural acceptability of the processed product. The extent to which the good texture can be achieved depends crucially on the quality of the raw materials [17]. This study suggests that there is no significant difference in hardness between two varieties of coconut, but there is a significant difference in the chewiness of two varieties of coconut. So this support the fact the texture of a food depends on the raw materials.

4. Conclusion

The antioxidant analysis states that antioxidant activity of fresh king coconut mesocarp was 360.1±26.4 mg/L and the antioxidant activity of fresh young coconut was 448.4±34.6 mg/L. But the antioxidant activity of standard Gallic acid was 4.3180±0.1171 mg/L, which has considerably higher antioxidant activity than of coconut mesocarp.

But among the mesocarps of tender king and young coconut, there is no significant difference in texture parameter "hardness" and there is a significant difference in texture parameter "chewiness" at 5% significant level. The hardness value and chewiness value of fresh king and young coconut mesocarps were 1306.3±44.6 & 24.92±2.06 and 1281.3±70.8 & 79.62±5.56 respectively.

Tender king coconut mesocarp has higher antioxidant activity and lower chewiness than tender young coconut mesocarp. So based on antioxidant activity and texture profile, king coconut mesocarp is a good source in developing healthy food product. The analysis of present study suggests that it is good practice to develop products in such a way that enhances the antioxidant quality.

5. References

1. Aragao WM. Côco: pós-colheita. Série frutas do Brasil. Brasília: Embrapa Informação Tecnológica, 2002.
2. Purseglove JW. Tropical crops: monocotyledons. London: Longman, 1972.
3. Dardick C, Callahan AM. Evolution of the fruit endocarp: molecular mechanisms underlying adaptations in seed protection and dispersal strategies. *Front Plant Sci.* 2014; 5:1-10.
4. Passos EEM. Morfologia do coqueiro. A cultura do coqueiro no Brasil. 2nd edn. Brasília: Embrapa - Serviço de Produção de Informação, 1998.
5. Jarimopas B, Ruttanadat N. Development of a young coconut fruit trimming machine. *Journal of Food Engineering.* 2007; 79:752-757.
6. Gatchalian MM, De Leon SY, Yano T. Measurement of young coconut (*Cocos nucifera*, L.) maturity by sound waves. *Journal of Food Engineering.* 1994; 23:253-276.
7. Lima EBC *et al.* *Cocos nucifera* (L.) (Arecaceae): A phytochemical and pharmacological review. *Braz J Med Biol Res [online].* 2015; 48(11):953-964.
8. Appaiah P, LS, AG, GK, GSK. Phytochemicals and Antioxidant Activity of Testa Extracts of Commercial Wet and Dry Coconuts and Cakes. *International Research Journal of Pharmacy.* 2016; 7(9):9-13.
9. Peuchant Brun E, Rigalleau J, Dubourg V, Thomas L, Daniel M. J Oxidative and antioxidative status in pregnant women with either gestational or type 1 diabetes. *Clin Biochem.* 2004; 37:293-298.
10. Brand-Williams W, Cuvelier ME, Berset C. Use of a free radical method to evaluate antioxidant activity. *J Food Sci Technol.* 1995; 28:25-30.
11. Thomson RL, Fleming HP, Hamann DD. Delineation of puncture forces for exocarp and mesocarp tissues in cucumber fruit. *Journal of texture studies.* 1992; 23:169-184.
12. Wasala WMCB, Thilakarathne BMKS. Retention of physicochemical and antioxidant properties of dehydrated bael (*Aegle marmelos*) and palmyra (*Borassus flabellifer*) fruit powders. *Italian Oral Surgery.* 2016; 6(2015):170-175.
13. Venkatesh R, Sood D. A Review of the Physiological Implications of Antioxidants in Food. *A Review of the Physiological Implications of Antioxidants in Food.* 2011.
14. Prior RL, Wu X, Schaich K. Standardized methods for the determination of antioxidant activity and phenolics in foods and dietary supplements. *J Agri Food Chem.* 2005; 53:4290-4302.

15. Blois MS. Antioxidant determination by the case of a stable free radical, *Nature*, 1958; 181:1199-1200.
16. Sultana B, Anwas F, Iqbal S. Effect of different cooking methods on the antioxidant activity of some vegetables from Pakistan, *Int J Food Sci Technol* doi, 2007, 1365-2621.
17. Adams JB. (n.d.). Raw materials quality and the texture of processed vegetables. *Texture in food*. Wood head Publishing Limited, 2004. <https://doi.org/10.1533/978185538362.3.342>.